



Europäisches Patentamt
European Patent Office
Office européen des brevets



(11) Publication number:

0 601 322 A2

(12)

EUROPEAN PATENT APPLICATION

(21) Application number: 93117408.0

(5) Int. Cl.5: A61K 31/70, C07H 19/16

(22) Date of filing: 27.10.93

(30) Priority: 27.10.92 JP 312764/92
27.10.92 JP 312765/92

(43) Date of publication of application:
15.06.94 Bulletin 94/24

(64) Designated Contracting States:
AT BE CH DE DK ES FR GB GR IE IT LI LU MC
NL PT SE

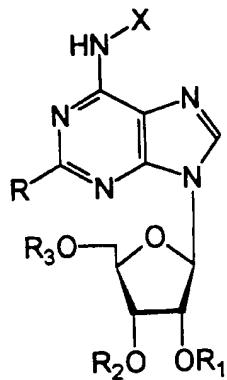
(71) Applicant: NIPPON ZOKI PHARMACEUTICAL
CO., LTD.
1-2, Hiranomachi Nichome
Chuo-ku, Osaka(JP)

(72) Inventor: Yamada, Toshio, c/o Inst. of
Bio-Active Science
Nippon Zoki Pharma. Co.,Ltd.,
442-1, Kinashi
Yashiro-cho, Katoh-gun, Hyogo(JP)

(74) Representative: Hansen, Bernd, Dr.
Dipl.-Chem. et al
Hoffmann, Eitlie & Partner,
Patentanwälte,
Arabellastrasse 4
D-81925 München (DE)

(54) Adenosine deaminase inhibitor.

(57) The use of a compound represented by the following general formula (I) or a pharmaceutically acceptable salt thereof



(I)

EP 0 601 322 A2

wherein each of R₁, R₂ and R₃ which may be same or different is hydrogen or alkyl; and R and X have the meanings given in the specification, for the preparation of a medicament which is an adenosine deaminase inhibitor is disclosed. Some of the compounds of formula (I) are novel compounds. They may be used for the making of a medicament which is for the prevention or therapy of ischemic heart diseases, diseases caused by cerebrovascular disorders, renal diseases and allergic diseases and of post-operative complicated diseases involved with activated oxygen which is generated in ischemic areas during the recirculation of blood after operations.

BEST AVAILABLE COPY

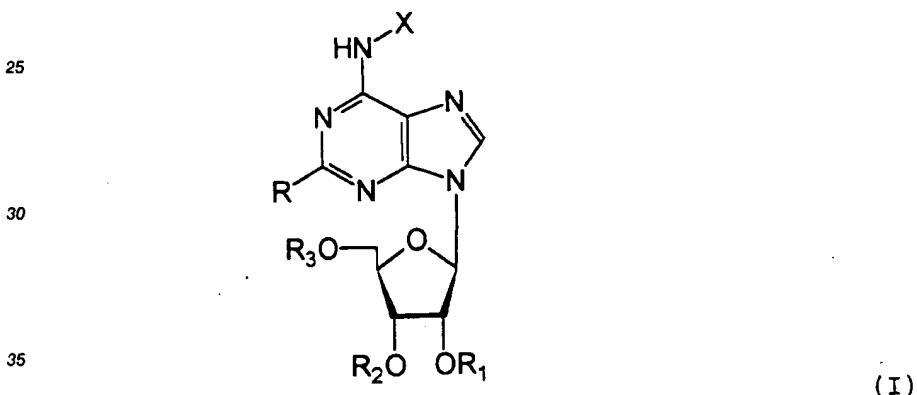
The present invention relates to adenosinedeaminase inhibitors containing at least one of certain adenosine derivatives and pharmaceutically acceptable salts thereof as an effective component.

Adenosinedeaminase is an enzyme producing inosine by deamination of adenosine in vivo and is widely distributed throughout animals and microorganisms. When said adenosinedeaminase is inhibited, adenosine concentration in tissues is increased while inosine concentration decreases whereupon endogenous inactivation of adenosine is inhibited. When the tissue is in an ischemic state, neutrophils produce activated oxygen and adenosine inhibits said oxygen production and, in addition, it has an action of direct elimination of the produced activated oxygen. Further, as a result of the decrease in the inosine concentration, supply of hypoxanthine which is a substrate in the xanthine-xanthineoxidase system (which is one of the systems producing activated oxygen) is decreased. It has been known that adenosinedeaminase inhibitors which inhibit the production of such activated oxygen sources and also eliminate them exhibit pharmacological actions such as improvement of coronary and cerebral blood vessel circulation, prevention and therapy of renal diseases, anti-inflammatory activity, etc.

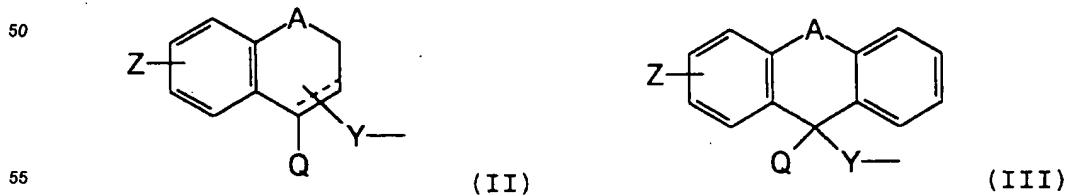
The present inventor has conducted investigations on O-alkylated adenosine derivatives, found that the compounds of the present invention exhibit excellent adenosinedeaminase inhibiting action and accomplished the present invention.

One object of the present invention is to provide a novel use of certain adenosine derivatives. A further object of the present invention is to offer a pharmaceutical composition containing at least one of certain adenosine derivatives having adenosinedeaminase inhibiting action as an effective component.

The present invention relates to the use of a compound represented by the following general formula (I) or a pharmaceutically acceptable salt thereof



wherein each of R₁, R₂ and R₃ which may be same or different is hydrogen or alkyl;
 40 R is hydrogen, alkyl, alkenyl, alkynyl, hydroxyalkynyl, alkoxy, phenyl, hydroxy, amino, alkylamino, phenylamino or halogen;
 X is hydrogen, alkyl, alkynyl, allyl, methallyl, cycloalkyl, an alkyl group being substituted with one or more hydroxy groups, phenyl (which may be substituted with one or more halogen atoms, lower alkyl groups, lower alkoxy groups and/or trifluoromethyl groups), an alkyl group being substituted with one or more phenyl groups (which may be substituted with one or more halogen atoms, lower alkyl groups, lower alkoxy groups and/or trifluoromethyl groups), bicycloalkyl, naphthylalkyl, acenaphthylenealkyl or a group represented by the following general formulae (II) or (III)



wherein Z is hydrogen, hydroxy or lower alkoxy;

Q is hydrogen or hydroxy;

A is $-\text{CH}_2-$, $-\text{O}-$, $-\text{S}-$ or a single bond;

Y is $-(\text{CH}_2)_n-$ or a single bond with n being an integer of 1 to 3;

5 a dashed line means a single bond or a double bond;

and wherein at least one of R_1 , R_2 and R_3 is alkyl,

for the preparation of a medicament which is an adenosinedeaminase inhibitor.

In the above general formula (I), each of R_1 , R_2 and R_3 may be same or different and is hydrogen or alkyl. Preferably, R_1 is hydrogen or a linear or branched alkyl group having 1 to 10 carbon atoms such as

10 methyl, ethyl, propyl, isopropyl, butyl, isobutyl, sec-butyl, tert-butyl, pentyl, isopentyl, neopentyl, tert-pentyl, hexyl, dimethylbutyl, heptyl, octyl, nonyl and decyl, and R_2 and R_3 are hydrogen or a linear or branched alkyl group having 1 to 4 carbon atoms such as methyl, ethyl, propyl, isopropyl, butyl, isobutyl, sec-butyl and tert-butyl. Most preferably, R_3 is hydrogen.

R is hydrogen; alkyl, preferably a linear or branched alkyl group having 1 to 20 carbon atoms such as

15 methyl, ethyl, propyl, isopropyl, butyl, isobutyl, sec-butyl, tert-butyl, pentyl, isopentyl, neopentyl, tert-pentyl, hexyl, dimethylbutyl, heptyl, octyl, nonyl, decyl and stearyl; alkenyl, preferably a linear or branched alkenyl group having 2 to 4 carbon atoms such as ethenyl, 1-propenyl, 2-propenyl, 1-butenyl, 2-butenyl and 3-but enyl; alkynyl, preferably a linear or branched alkynyl group having 2 to 20 carbon atoms such as ethynyl, 1-propynyl, 2-propynyl, 1-butynyl, 2-butynyl, 3-butynyl, sec-butynyl, pentynyl, isopentynyl, 2-

20 pentynyl, 3-pentynyl, 4-pentynyl, hexynyl, heptynyl, octynyl, nonynyl, decynyl and stearynyl; hydroxylkynyl, preferably one of the above alkynyl groups substituted with one or more hydroxy groups; alkoxy, preferably a linear or branched alkoxy group having 1 to 4 carbon atoms such as methoxy, ethoxy, propoxy, isoproxy, butoxy, isobutoxy, sec-butoxy and tert-butoxy; phenyl; hydroxy; amino; alkylamino, preferably a linear or branched mono- or dialkylamino group having 1 to 10 carbon atoms such as methylamino,

25 dimethylamino, ethylamino, diethylamino, methylethylamino, propylamino, isopropylamino butylamino, isobutylamino, sec-butylamino, tert-butylamino, pentylamino, isopentylamino, neopentylamino, tert-pentylamino, hexylamino, dimethylbutylamino, heptylamino, octylamino, nonylamino and decylamino; phenylamino; or halogen such as fluorine, chlorine, bromine and iodine. In those symbols, at least one of R_1 , R_2 and R_3 is to represent alkyl.

30 X is hydrogen; alkyl, preferably a linear or branched alkyl group having 1 to 6 carbon atoms such as methyl, ethyl, propyl, isopropyl, butyl, isobutyl, sec-butyl, tert-butyl, pentyl, isopentyl, neopentyl, tert-pentyl, hexyl and dimethylbutyl; alkynyl, preferably a linear or branched alkynyl group having 2 to 7 carbon atoms such as ethynyl, propynyl, butynyl, pentynyl, hexynyl and heptynyl; allyl; methallyl; cycloalkyl, preferably a cycloalkyl group having 3 to 8 carbons such as cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, cycloheptyl

35 and cyclooctyl; an alkyl group being substituted with one or more hydroxy groups, preferably a linear or branched alkyl group having 1 to 4 carbon atoms such as methyl, ethyl, propyl, isopropyl, butyl, isobutyl, sec-butyl and tert-butyl substituted with one or two hydroxy groups; phenyl (which may be substituted with one or more halogen atoms, lower alkyl groups, lower alkoxy groups and/or trifluoromethyl groups); an alkyl group being substituted with one or more phenyl groups (which may be substituted with one or more

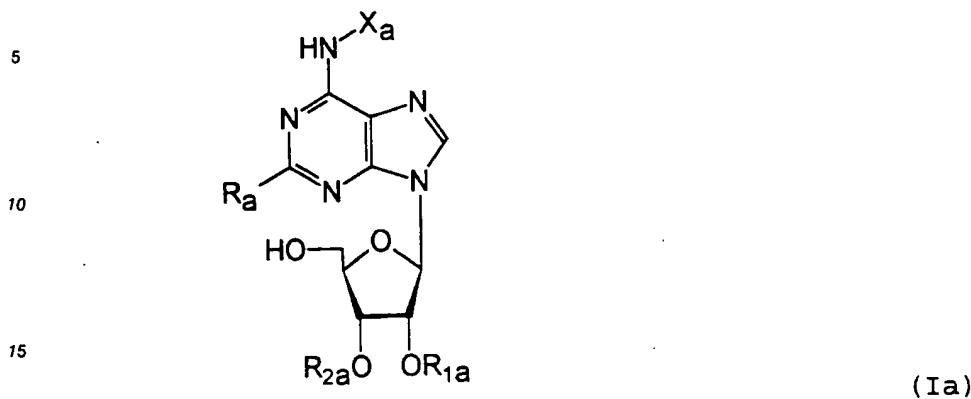
40 halogen atoms, lower alkyl groups having 1-6 carbon atoms, lower alkoxy groups having 1-6 carbon atoms and/or trifluoromethyl groups), preferably a phenylalkyl or diphenylalkyl group in which one or two optionally substituted phenyl groups as given above is/are bonded to a linear or branched alkyl group having 1 to 3 carbon atoms such as methyl, ethyl, propyl and isopropyl; a bicycloalkyl group, preferably endo- or exo-bicyclo[2.2.1]heptyl; a naphthylalkyl group, wherein preferably an optionally substituted naphthyl group is

45 bonded to an alkyl group having 1 to 3 carbon atoms such as methyl, ethyl, propyl and isopropyl; acenaphthylenealkyl (including its 1,2-dihydro form), preferably an acenaphthylenealkyl group in which acenaphthylene is bonded to an alkyl group having 1 to 3 carbon atoms such as methyl, ethyl, propyl and isopropyl; or a group represented by the above-mentioned general formula (II) or (III). It is particularly preferable that X is hydrogen or alkyl having 1 to 3 carbon atoms.

50 In the general formulae (II) and (III), Z is hydrogen, hydroxy or a lower alkoxy group, preferably a linear or branched alkoxy group having 1 to 3 carbon atoms such as methoxy, ethoxy, propoxy and isopropoxy; Q is hydrogen or hydroxy; A is $-\text{CH}_2-$, $-\text{O}-$, $-\text{S}-$ or a single bond; Y is $-(\text{CH}_2)_n-$ or a single bond; n is an integer of 1 to 3; and a dashed line means a single bond or a double bond.

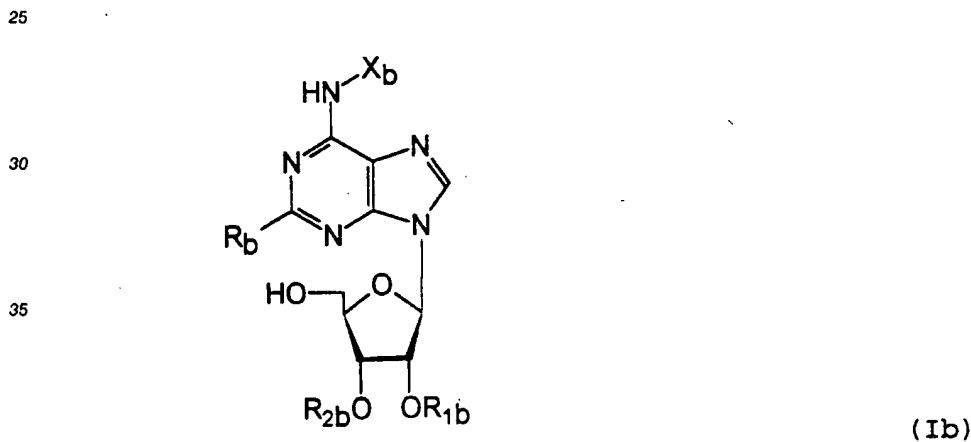
The said adenosine derivatives of the present invention include novel compounds as follows:

(1) Adenosine derivatives represented by the following general formula (Ia):



20 wherein each of R_{1a} , R_{2a} and X_a may be same or different and is hydrogen or alkyl; R_a is alkyl having more than 6 carbon atoms, alkenyl, alkynyl, hydroxyalkynyl, alkoxy, phenyl, hydroxy, alkylamino or phenylamino; and at least one of R_{1a} and R_{2a} is alkyl.

(2) Adenosine derivatives represented by the following general formula (Ib)

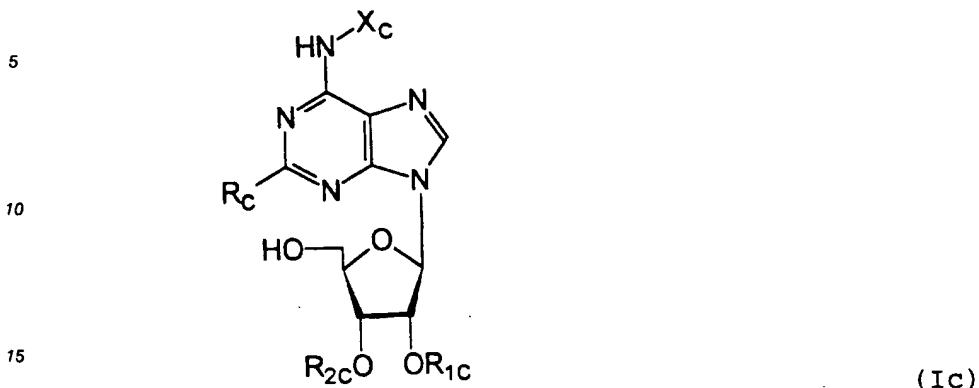


45 wherein each of R_{1b} and R_{2b} may be same or different and is alkyl; R_b and X_b is hydrogen or alkyl; and at least one of R_b and X_b is alkyl.

50

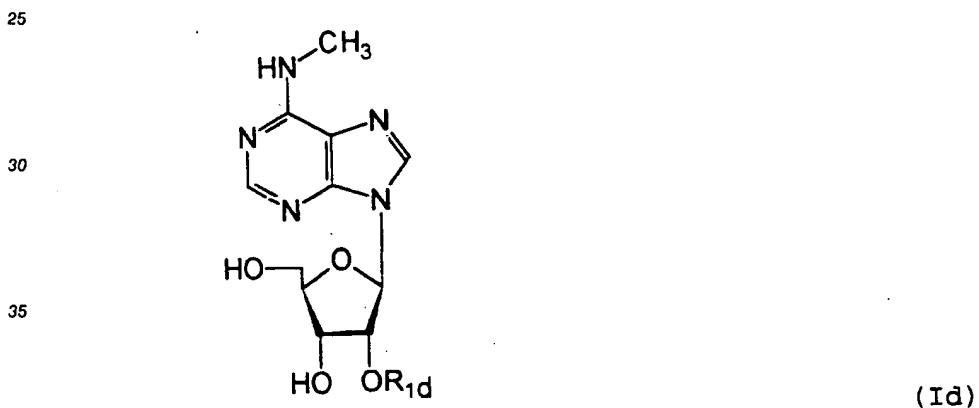
55

(3) Adenosine derivatives represented by the following general formula (Ic)



20 wherein each of R_{1c}, R_{2c} and X_c may be same or different and is hydrogen or alkyl ; R_c is bromo or iodo; and at least one of R_{1c} and R_{2c} is alkyl.

(4) Adenosine derivatives represented by the following general formula (Id)



40

wherein R_{1d} is alkyl having more than 2 carbon atoms.

Preferred substituents R_a, R_b, R_{1a}, R_{1b}, R_{2a} etc. in the above adenosine derivatives represented by formulae (Ia) to (Id) are the same groups as indicated under formula (I).

45 Examples of compounds which may be contained as an effective ingredient in the adenosinedeaminase inhibitor in accordance with the present invention are as follows:

- (Compound 1) 2'-O-Methyladenosine;
- (Compound 2) 3'-O-Methyladenosine;
- (Compound 3) 2'-O-Ethyladenosine;
- 50 (Compound 4) 2'-O-n-Butyladenosine;
- (Compound 5) 2,2'-O-Dimethyladenosine;
- (Compound 6) 2,3'-O-Dimethyladenosine;
- (Compound 7) 2-Isopropyl-2'-O-methyladenosine;
- (Compound 8) 2-Isopropyl-3'-O-methyladenosine
- 55 (Compound 9) 2-Methoxy-3'-O-methyladenosine
- (Compound 10) 2-Methyl-2'-O-ethyladenosine
- (Compound 11) 2-Methyl-2'-O-n-butyladenosine
- (Compound 12) 5'-O-Methyladenosine

- (Compound 13) 5'-O-n-Butyladenosine
- (Compound 14) 2',5'-O-Dimethyladenosine
- (Compound 15) 3',5'-O-Dimethyladenosine
- (Compound 16) 2,5'-O-Dimethyladenosine
- 5 (Compound 17) 2-Methyl-5'-O-n-butyladenosine
- (Compound 18) N⁶,2'-O-Dimethyladenosine
- (Compound 19) N⁶-Ethyl-2'-O-methyladenosine
- (Compound 20) N⁶-n-Butyl-2'-O-methyladenosine
- (Compound 21) N⁶-Methyl-2'-O-ethyladenosine
- 10 (Compound 22) N⁶-Methyl-2'-O-n-butyladenosine
- (Compound 23) N⁶-Methyl-2'-O-n-hexyladenosine
- (Compound 24) N⁶-Methyl-2'-O-n-octyladenosine
- (Compound 25) N⁶,5'-O-Dimethyladenosine
- (Compound 26) N⁶-n-Butyl-5'-O-methyladenosine
- 15 (Compound 27) 2,N⁶,2'-O-Trimethyladenosine
- (Compound 28) 2,N⁶-Dimethyl-2'-O-ethyladenosine
- (Compound 29) N⁶-n-Butyl-2,2'-O-dimethyladenosine
- (Compound 30) N⁶,2'-O-Dimethyl-2-hexyladenosine
- (Compound 31) N⁶,2'-O-Dimethyl-2-decyadenosine
- 20 (Compound 32) N⁶,2'-O-Dimethyl-2-(1-hexyn-1-yl)-adenosine
- (Compound 33) N⁶,2'-O-Dimethyl-2-(1-dodecyn-1-yl)-adenosine
- (Compound 34) 2,N⁶,5'-O-Trimethyladenosine
- (Compound 35) N⁶-n-Butyl-2,5'-O-dimethyladenosine
- (Compound 36) 2,N⁶,3'-O-Trimethyladenosine
- 25 (Compound 37) 2-Phenyl-2'-O-methyladenosine
- (Compound 38) 2-Phenyl-3'-O-methyladenosine
- (Compound 39) 2-Hydroxy-2'-O-methyladenosine
- (Compound 40) 2-Hydroxy-3'-O-methyladenosine
- (Compound 41) 2-Chloro-2'-O-methyladenosine
- 30 (Compound 42) 2-Chloro-3'-O-methyladenosine
- (Compound 43) 2-Bromo-2'-O-methyladenosine
- (Compound 44) 2-Bromo-3'-O-methyladenosine
- (Compound 45) 2-Bromo-N⁶,2'-O-dimethyladenosine
- (Compound 46) 2-Bromo-N⁶,3'-O-dimethyladenosine
- 35 (Compound 47) 2-Iodo-2'-O-methyladenosine
- (Compound 48) 2-Iodo-3'-O-methyladenosine
- (Compound 49) 2-Fluoro-2'-O-methyladenosine
- (Compound 50) 2-Amino-2'-O-methyladenosine
- (Compound 51) 2-Amino-3'-O-methyladenosine
- 40 (Compound 52) 2-Pentylamino-2'-O-methyladenosine
- (Compound 53) 2-Phenylamino-2'-O-methyladenosine
- (Compound 54) 2-Phenylamino-3'-O-methyladenosine
- (Compound 55) 2-Phenylamino-N⁶,2'-O-dimethyladenosine
- (Compound 56) 2-Phenylamino-N⁶,3'-O-dimethyladenosine
- 45 (Compound 57) 2-(3-Hydroxy-1-propyn-1-yl)-2'-O-methyladenosine
- (Compound 58) 2-(3-Hydroxy-1-propyn-1-yl)-3'-O-methyladenosine
- (Compound 59) 2',3'-O-Dimethyladenosine
- (Compound 60) N⁶,2',3'-O-Trimethyladenosine
- (Compound 61) N⁶-Methyl-2',3'-O-diethyladenosine
- 50 (Compound 62) N⁶-n-Butyl-2',3'-O-dimethyladenosine
- (Compound 63) 2,2',3'-O-Trimethyladenosine
- (Compound 64) 2,N⁶,2',3'-O-Tetramethyladenosine
- (Compound 65) N⁶-Allyl-2'-O-methyladenosine
- (Compound 66) N⁶-Methallyl-2'-O-methyladenosine
- 55 (Compound 67) N⁶-(2,3-Dihydroxypropyl)-2'-O-methyladenosine
- (Compound 68) N⁶-Cyclopropyl-2'-O-methyladenosine
- (Compound 69) N⁶-Cyclopentyl-2'-O-methyladenosine
- (Compound 70) N⁶-Cyclopentyl-2'-O-ethyladenosine

- (Compound 71) N⁶-Cyclopentyl-2',2'-dimethyladenosine
- (Compound 72) N⁶-Cyclopentyl-2-bromo-2'-O-methyl- adenosine
- (Compound 73) N⁶-Cyclohexyl-2'-O-methyladenosine
- (Compound 74) N⁶-Cyclohexyl-2',2'-O-dimethyladenosine
- 5 (Compound 75) N⁶-Cycloheptyl-2'-O-methyladenosine
- (Compound 76) N⁶-p-Methoxyphenyl-2'-O-methyladenosine
- (Compound 77) N⁶-p-Fluorophenyl-2'-O-methyladenosine
- (Compound 78) N⁶-p-Chlorophenyl-2'-O-methyladenosine
- (Compound 79) N⁶-Benzyl-2'-O-methyladenosine
- 10 (Compound 80) N⁶-(R)-Phenyl-isopropyl-2'-O-methyladenosine
- (Compound 81) N⁶-(2,2-Diphenylethyl)-2'-O-methyl-adenosine
- (Compound 82) N⁶-(exo-Bicyclo[2.2.1]heptyl)-2'-O-methyladenosine
- (Compound 83) N⁶-(endo-Dicyclo[2.2.1]heptyl)-2'-O-methyladenosine
- (Compound 84) N⁶-(1-Naphthyl)methyl-2'-O-methyl-adenosine
- 15 (Compound 85) N⁶-1-(Acenaphthylene)ethyl-2'-O-methyladenosine
- (Compound 86) N⁶-(1,2-Dihydro-1-acenaphthylene)methyl-2'-O-methyladenosine
- (Compound 87) N⁶-(2,3-Dihydro-1H-inden-1-yl)-2'-O-methyladenosine
- (Compound 88) N⁶-(2,3-Dihydro-1H-inden-2-yl)-2'-O-methyladenosine
- (Compound 89) N⁶-(2,3-Dihydro-1H-inden-1-yl)methyl-2'-O-methyladenosine
- 20 (Compound 90) N⁶-(3H-Inden-1-yl)methyl-2'-O-methyladenosine
- (Compound 91) N⁶-(5-Methoxy-2,3-dihydro-1H-inden-2-yl)-2'-O-methyladenosine
- (Compound 92) N⁶-(1-Tetrahydronaphthyl)-2'-O-methyladenosine
- (Compound 93) N⁶-(2-Tetrahydronaphthyl)-2'-O-methyladenosine
- (Compound 94) N⁶-(3,4-Dihydro-1-naphthyl)methyl-2'-O-methyladenosine
- 25 (Compound 95) N⁶-(5-Hydroxy-1-tetrahydronaphthyl)-2'-O-methyladenosine
- (Compound 96) N⁶-(1-Hydroxy-1-tetrahydronaphthyl)- methyl2'-O-methyladenosine
- (Compound 97) N⁶-(5-Methoxyl-1-tetrahydronaphthyl)- 2'-O-methyladenosine
- (Compound 98) N⁶-(6-Methoxy-1-tetrahydronaphthyl)-2'- O-methyladenosine
- (Compound 99) N⁶-(7-Methoxy-1-tetrahydronaphthyl)-2'- O-methyladenosine
- 30 (Compound 100) N⁶-(4-Chromanyl)-2'-O-methyladenosine
- (Compound 101) N⁶-(4-Thiochromanyl)-2'-O-methyl- adenosine
- (Compound 102) N⁶-Fluorenyl-2'-O-methyladenosine
- (Compound 103) N⁶-(9-Fluorenyl)methyl-2'-O-methyladenosine
- (Compound 104) N⁶-(9-Hydroxy-9-fluorenyl)methyl-2'-O-methyladenosine and
- 35 (Compound 105) N⁶-(9-Xanthenyl)methyl-2'-O-methyladenosine.

The compounds of the present invention as given above may be prepared by a method disclosed, for example, in Japanese Laid Open (Kokai) 63/329,294, 02/184,696 and 02/218,689.

The adenosine derivatives of the present invention also include the salts of the compounds represented by the general formulae given above, preferably those being pharmaceutically acceptable. Examples of such salts are acid addition salts such as salts with hydrochloric acid, sulfuric acid, nitric acid, hydrobromic acid, phosphoric acid, perchloric acid, thiocyanic acid, boric acid, formic acid, acetic acid, haloacetic acid, propionic acid, glycolic acid, citric acid, tartaric acid, succinic acid, gluconic acid, lactic acid, malonic acid, fumaric acid, anthranilic acid, benzoic acid, cinnamic acid, p-toluenesulfonic acid, naphthalenesulfonic acid and sulfanilic acid and salts with alkali metals (e.g. sodium and potassium), alkaline earth metals (e.g. calcium and magnesium) and other metals (e.g. aluminum). Adenosine derivatives of the present invention include metal complexes thereof such as, for example, complexes with zinc, nickel, cobalt, copper and iron. Those salts and metal complexes may be manufactured from free adenosine derivatives of the present invention or may be transformed into each other.

When there are stereoisomers for the compounds of the present invention such as cis-trans isomers, optical isomers and conformational isomers, the present invention includes all of them.

The following descriptions serve as illustrative examples for the preparation of compounds of the present invention.

Example 1

55 10 g of 6-chloro-9-(3,5-O-TIPDS)- β -D-ribofuranosyl-9H-purine was dissolved in 50 ml of ethyl iodide, and silver oxide was added and stirred with heating. The reaction mixture was applied on a silica gel column, washed with benzene and then eluted with ethyl acetate/hexane. The eluate was concentrated to

dryness under reduced pressure. The residue was dissolved in benzene and a 40%(w/v) aqueous solution of monomethylamine was added thereto. After stirring overnight, the benzene layer was separated, washed with 1N HCl and brine, and a mixture of 1M tetra-n-butylammonium and tetrahydrofuran was added thereto. The reaction mixture was stirred for 30 minutes at room temperature, concentrated under reduced pressure, and then purified by silica gel column chromatography to give 2.1 g of N⁶-methyl-2'-O-ethyladenosine (Compound 21).

1H-NMR (D₂O) : 1.21(3H,t), 3.19(3H,s), 3.58(1H,m), 3.69(1H,m), 3.85(1H,dd), 3.93(1H,dd), 4.32(1H,m), 4.55(1H,dd), 4.63(1H,dd), 6.10(1H,dd), 8.25(1H,s), 8.30(1H,s)

In the same manner as mentioned above, the following compounds were obtained.

10 N⁶-Methyl-2'-O-n-butyladenosine (Compound 22)

1H-NMR (DMSO-d₆):

0.74(3H,t), 1.17(2H,m), 1.37(2H,m), 2.96(3H,s), 3.34(1H,m), 3.55(2H,m), 3.68(1H,m), 3.99(1H,m), 4.29(1H,m), 4.47(1H,m), 5.15(1H,d), 5.42(1H,m), 5.99(1H,d), 7.81(1H,s), 8.23(1H,s), 8.37(1H,s)

N⁶-Methyl-2'-O-n-hexyladenosine (Compound 23)

15 1H-NMR (CDCl₃):

0.86(3H,t), 1.17(8H,m), 1.40(2H,m), 3.21(3H,s), 3.34(1H,m), 3.48(1H,m), 3.75(1H,m), 3.97(1H,m), 4.36(1H,m), 4.53(1H,d), 4.82(1H,dd), 5.82(1H,d), 5.87(1H,s), 6.87(1H,dd), 7.76(1H,s), 8.37(1H,s)

N⁶-Methyl-2'-O-n-octyladenosine (Compound 24)

1H-NMR (CDCl₃):

20 0.86(3H,t), 1.16(8H,m), 1.24(2H,m), 1.41(2H,m), 3.21(3H,s), 3.33(1H,m), 3.49(1H,m), 3.76(1H,m), 3.97(1H,m), 4.36(1H,m), 4.53(1H,d), 4.82(1H,dd), 5.82(1H,d), 6.02(1H,s), 6.87(1H,dd), 7.77(1H,s), 8.37(1H,s)

Example 2

25 5.34 g of adenosine was dissolved in 80 ml of dimethylformamide and 800 mg of 60%(w/w) sodium hydride in mineral oil was added thereto. After stirring for 30 minutes in ice-cold water, 2.84 g of methyl iodide in 10 ml of dimethylformamide was added dropwise. The reaction mixture was stirred under cooling for 2 hours and concentrated to dryness under reduced pressure. The residue was dissolved in water and applied on a cation exchange column. The fraction eluted with a 10%(v/v) aqueous solution of methanol was collected, concentrated to dryness, and purified by silica gel column chromatography to give 1.36 g of 2',3'-O-dimethyladenosine.

m.p.: 180 °C

1H-NMR (DMSO-d₆):

3.32(3H,s), 3.40(3H,s), 3.57(1H,m), 3.70(1H,m), 4.09(2H,m), 4.54(1H,dd), 5.47(1H,dd), 6.00(1H,d), 7.35(2H,s,D₂O-Disappearance), 8.14(1H,s), 8.38(1H,s)

Example 3

40 2 g of the compound obtained in Example 2 and 2 ml of methyl iodide were dissolved in dimethylacetamide, and stirred overnight at room temperature. The reaction mixture was evaporated to dryness under reduced pressure and 10 ml of 2N sodium hydroxide solution was added thereto. The solution was refluxed for one hour with heating. After cooling to room temperature, the solution was neutralized with 2N HCl and applied on an Amberlite XAD-7 column. The column was washed with water and eluted with a 50%(v/v) aqueous solution of methanol. The eluate was concentrated to dryness under 45 reduced pressure and recrystallized from ethyl acetate to give 1.8 g of N⁶,2',3'-O-trimethyladenosine (Compound 60); m.p.: 165 °C

1H-NMR (DMSO-d₆): 2.96(3H,s), 3.31(3H,s), 3.40(3H,s), 3.57(1H,m), 3.70(1H,m), 4.09(2H,m), 4.54(1H,dd), 5.49(1H,dd), 6.01(1H,d), 7.83(1H,s,D₂O-Disappearance), 8.24(1H,s), 8.38(1H,s)

50 Example 4

55 14.3 g of 6-chloro-9-β-D-ribofuranosyl-9H-purine and 15 g of triphenylchlorosilane were dissolved in 500 ml of pyridine and stirred for one hour at room temperature. Pyridine was distilled off and the residue was dissolved in benzene. The benzene layer was washed with 1N HCl and brine, and then dried over anhydrous sodium sulfate. The solvent was distilled off and the residue was recrystallized from a mixture of hexane and ethyl acetate to give 21.5 g of 6-chloro-9-(5-O-triphenylsilyl-β-D-ribofuranosyl)-9H-purine.

5.45 g of the resulting product was dissolved in 50 ml of ethyl iodide and silver oxide was added thereto with heating. The reaction mixture was applied on a silica gel column. The column was washed with

benzene and eluted with a mixture of hexane and ethyl acetate. The eluate was concentrated to dryness under reduced pressure. The residue was dissolved in benzene and a 40%(w/v) aqueous solution of monomethylamine was added thereto. After stirring overnight, the benzene layer was collected, washed with 1N HCl and brine and purified by silica gel column chromatography to give 6-amino-9-(5-O-triphenylsilyl- β -D-ribofuranosyl)-9H-purine. The resulting product was dissolved in tetrahydrofuran and tetra-n-butylammonium fluoride in tetrahydrofuran was added thereto. After stirring for 30 minutes at room temperature, the solution was concentrated to dryness under reduced pressure and purified by silica gel column chromatography to give 600 mg of N⁶-methyl- 2',3'-O-diethyladenosine (Compound 61). m.p.: amorphous
 1H-NMR (MeOH-d₄):
 10 1.07(3H,t), 1.22(3H,t), 3.08(3H,s), 3.48(1H,m), 3.58(1H,m), 3.65(2H,m), 3.70(1H,dd), 3.87(2H,m), 4.20(1H,dd), 6.00(1H,d), 8.20(1H,s), 8.23(1H,s)

Example 5

15 2-Methyladenosine was dialkylated in the same manner as in Example 2 to give 2,2',3'-O-trimethyladenosine (Compound 63). m.p.: 156 °C
 1H-NMR (DMSO-d₆): 2.39(3H,s), 3.28(3H,s), 3.40(3H,s), 3.58 (1H,m), 3.70(1H,m), 4.08(1H,m), 4.13(1H,m), 4.52(1H,m), 5.80(1H,m,D₂O-Disappearance), 5.95(1H,d), 7.27(2H,s,D₂O-Disappearance), 8.28(1H,s)

Example 6

Compound 63 was methylated by methyl iodide and then rearranged to give 2,N⁶,2',3'-O-tetramethyladenosine (Compound 64).
 1H-NMR (DMSO-d₆):

25 2.43(3H,s), 2.94(3H,s), 3.28(3H,s), 3.40(3H,s), 3.57(1H,m), 3.70(1H,m), 4.08(1H,m), 4.13(1H,m), 4.52(1H,m), 5.79(1H,m,D₂O-Disappearance), 5.96(1H,d), 7.71(1H,s,D₂O-Disappearance), 8.26(1H,s)

Example 7

30 1.2 g of 2-iodoadenosine was suspended in 150 ml of 1 mmol tin chloride dihydrate/methanol. 50 ml of 0.4-0.5M diazomethane in 1,2-dimethoxyethane was added under stirring. After stirring for one hour at room temperature, the reaction mixture was concentrated to dryness under reduced pressure. The resulting product was applied on an ODS column and eluted with 40%(v/v) methanol in 0.1%(v/v) aqueous solution of TFA. First, 2-iodo-2'-O-methyladenosine (Compound 47) was eluted, and then 2-iodo-3'-O-methyladenosine (Compound 48) was eluted. Both fractions were concentrated to dryness to give 135 mg of compound 47 and 56 mg of Compound 48.

2-iodo-2'-O-methyladenosine (Compound 47)

1H-NMR (D₂O): 3.52(3H,s), 3.92(1H,dd), 3.97(1H,dd), 4.27(1H,m), 4.54(1H,dd), 4.61(1H,dd), 6.02(1H,d), 7.98(1H,s)

2-iodo-3'-O-methyladenosine (Compound 48)

1H-NMR (D₂O): 3.58(3H,s), 3.86(1H,dd), 3.94 (1H,dd), 4.18(1H,dd), 4.35(1H,m), 4.97(1H,dd), 5.93(1H,d), 7.94(1H,s)

In the same manner by using 2-substituted adenosines, N⁶-substituted adenosines or 2,N⁶-disubstituted adenosines, the following compounds were obtained.

45 2-Methoxy-3'-O-methyladenosine (Compound 9)

1H-NMR (D₂O): 3.56(3H,s), 3.84(1H,dd), 3.94(1H,dd), 3.96(3H,s), 4.22(1H,dd), 4.33(1H,m), 5.06(1H,dd), 5.98-(1H,d), 8.10(1H,s,8-H)

N⁶,2'-O-Dimethyl-2-hexyladenosine (Compound 30)

1H-NMR (MeOH-d₄): 0.88(3H,t), 1.33(6H,m), 1.78(2H,quintet), 2.74(2H,t), 3.12(3H,brs), 3.37(3H,s), 3.73-(1H,dd), 3.88(1H,dd), 4.18(1H,m), 4.49(2H,m), 5.98(1H,d), 8.10(1H,s,8-H)

N⁶,2'-O-Dimethyl-2-decyladenosine (Compound 31)

1H-NMR (MeOH-d₄): 0.88(3H,t), 1.27-1.33(14H,m), 1.78(2H,quintet), 2.75(2H,t), 3.13(3H,brs), 3.37(3H,s), 3.73(1H,dd), 3.89(1H,dd), 4.18(1H,m), 4.49(2H,m)

N⁶,2'-O-Dimethyl-2-(1-hexyn-1-yl)adenosine (Compound 32)

1H-NMR (MeOH-d₄): 0.97(3H,t), 1.52(2H,m), 1.62(2H,m), 2.45(2H,t), 3.09(3H,brs), 3.40(3H,s), 3.74(1H,dd), 3.88(1H,dd), 4.15(1H,m), 4.42(1H,dd), 4.48(1H,dd), 6.00(1H,d), 8.22(1H,s)

N⁶,2'-O-Dimethyl-2-(1-dodecyn-1-yl)adenosine (Compound 33)

1H-NMR (MeOH-d₄): 0.89(3H,t), 1.30(14H,m), 1.49(2H,m), 1.63(2H,quintet), 2.43(2H,t), 3.41(3H,s), 3.74-

(1H,dd), 3.88(1H,dd), 4.15(1H,m), 4.41(1H,dd), 4.48(1H,dd), 6.00(1H,d), 8.30(1H,s)

2-Phenyl-2'-O-methyladenosine (Compound 37)
 1H-NMR (MeOH-d₄): 3.53(3H,s), 3.77(1H,dd), 3.91(1H,dd), 4.12(1H,m), 4.50(1H,dd), 4.54(1H,dd), 6.20(1H,d), 7.43(3H,m), 8.32(2H,m), 8.36(1H,s)

5 2-Phenyl-3'-O-methyladenosine (Compound 38)
 1H-NMR (MeOH-d₄): 3.53(3H,s), 3.74(1H,dd), 3.88(1H,dd), 4.10(1H,dd), 4.22(1H,m), 5.00(1H,dd), 6.07(1H,d), 7.42(3H,m), 8.32(3H,m)

2-Bromo-2'-O-methyladenosine (Compound 43)
 1H-NMR (D₂O): 3.50(3H,s), 3.91(1H,dd), 3.99(1H,dd), 4.37(1H,m), 4.57(1H,dd), 4.67(1H,dd), 6.13(1H,d), 8.37(1H,s)

10 2-Bromo-3'-O-methyladenosine (Compound 44)
 1H-NMR (D₂O): 3.60(3H,s), 3.90(1H,dd), 4.01(1H,dd), 4.19(1H,dd), 4.44(1H,m), 4.95(1H,dd), 6.07(1H,d), 8.37(1H,s)

2-Bromo-N⁶,2'-O-dimethyladenosine (Compound 45)
 1H-NMR (D₂O): 3.22(3H,s), 3.57(3H,s), 3.90(1H,dd), 3.98(1H,dd), 4.29(1H,m), 4.59(1H,dd), 4.65(1H,dd), 6.12(1H,d), 8.16(1H,s)

15 2-Bromo-N⁶,3'-O-dimethyladenosine (Compound 46)
 1H-NMR (D₂O): 3.07(3H,s), 3.60(3H,s), 3.93(1H,dd), 3.99(1H,dd), 4.23(1H,dd), 4.36(1H,m), 5.03(1H,dd), 6.03(1H,d), 8.14(1H,s)

20 2-Pentylamino-2'-O-methyladenosine (Compound 52)
 1H-NMR (MeOH-d₄): 0.94(3H,t), 1.38(4H,m), 1.65(2H,m), 3.42(2H,t), 3.49(3H,s), 3.76(1H,dd), 3.87(1H,dd), 4.08(1H,m), 4.25(1H,t), 4.43(1H,t), 5.99(1H,d), 8.22(1H,s)

2-Phenylamino-2'-O-methyladenosine (Compound 53)
 1H-NMR (MeOH-d₄): 3.46(3H,s), 3.74(1H,dd), 3.85(1H,dd), 4.07(1H,m), 4.23(1H,dd), 4.39(1H,dd), 6.03(1H,d), 7.11(1H,t), 7.35(2H,t), 7.60(2H,d), 8.35(1H,s)

25 2-Phenylamino-3'-O-methyladenosine (Compound 54)
 1H-NMR (MeOH-d₄): 3.46(3H,s), 3.69(1H,dd), 3.79(1H,dd), 3.94(1H,dd), 4.16(1H,m), 4.74(1H,dd), 5.91(1H,d), 7.13(1H,t), 7.36(2H,t), 7.60(2H,d), 8.28(1H,s)

2-Phenylamino-N⁶,2'-O-dimethyladenosine (Compound 55)
 1H-NMR (MeOH-d₄): 3.16(3H,s), 3.47(3H,s), 3.77(1H,dd), 3.88(1H,dd), 4.11(1H,m), 4.20(1H,dd), 4.40(1H,dd), 6.06(1H,d), 7.12(1H,t), 7.35(2H,t), 7.63(2H,d), 8.42(1H,s)

30 2-Phenylamino-N⁶,3'-O-dimethyladenosine (Compound 56)
 1H-NMR (MeOH-d₄): 3.10(3H,s), 3.49(3H,s), 3.72(1H,dd), 3.85(1H,dd), 3.98(1H,dd), 4.17(1H,m), 4.85(1H,dd), 5.88(1H,d), 6.94(1H,t), 7.26(2H,t), 7.69(2H,d), 7.99(1H,s)

35 2-(3-Hydroxy-1-propyn-1-yl)-2'-O-methyladenosine (Compound 57)
 1H-NMR (D₂O): 3.45(3H,s), 3.86(1H,dd), 3.95(1H,dd), 4.32(1H,m), 4.51(1H,dd), 4.52(2H,s), 4.62(1H,dd), 6.08(1H,d), 8.30(1H,s)

2-(3-Hydroxy-1-propyn-1-yl)-3'-O-methyladenosine (Compound 58)
 1H-NMR (D₂O + MeOH-d₄): 3.51(3H,s), 3.80(1H,dd), 3.92(1H,dd), 4.09(1H,m), 4.36(1H,m), 4.47(2H,s), 4.83(1H,dd), 5.96(1H,d), 8.30(1H,s)

40

Example 8

45 2.2 g of AICA-2'-O-methylriboside was dissolved in 20 ml of dimethylformamide and 1.7 g of benzoylisothiocyanate was added thereto. After stirring for 3 hours at room temperature, 3.3 g of dicyclohexylcarbodiimide was added and reacted for 20 hours at room temperature. The reaction mixture was concentrated to an oily residue. To the residue, 50 ml of ethanol and 50 ml of aqua ammonia were added, and stirred for 18 hours at room temperature. The resulting precipitate was collected by filtration to give 1.7 g of 2-hydroxy-2'-O-methyladenosine (Compound 39). 1H-NMR (D₂O): 3.46(3H,s), 3.82(1H,dd), 3.90(1H,dd), 4.27(1H,m), 4.50(1H,dd), 4.59(1H,dd), 5.95(1H,d), 8.03(1H,s)

50 In the same manner, AICA-3'-O-methylriboside was used as a starting material to give 2-hydroxy-3'-O-methyladenosine (Compound 40).
 1H-NMR (D₂O): 3.54(3H,s), 3.82(1H,dd), 3.94(1H,dd), 4.10(1H,dd), 4.36(1H,m), 4.86(1H,dd), 5.89(1H,dd), 8.03(1H,s)

55 The following description serves to illustrate the pharmaceutical activity of the compounds of the present invention.

1. Adenosinedeaminase Inhibiting Action.

The enzymatic reaction was conducted at 25°C in a 0.05M phosphate buffer (pH=7.5). Thus, a reaction solution comprising 800 µl of substrate solution (adenosine), 100 µl of a solution to be tested and 5 100 µl of an enzyme solution (adenosinedeaminase of type VII mucous membrane of the calf intestinal tract) was made to react for 5 minutes and the reaction was stopped by adding 100 µl of acetic acid. Then the amount of inosine produced was determined by means of HPLC to measure the inhibiting activity. The test was conducted for various substrate concentrations and Ki values were measured by Lineweaver-Burk plots.

10 Examples of the results are given in Table 1.

TABLE 1

15	Compound tested	Ki Value (M)	Compound tested	Ki Value (M)
18	1	1.31×10^{-5}	39	1.90×10^{-5}
20	2	4.93×10^{-4}	43	3.20×10^{-6}
22	4	3.47×10^{-6}	44	2.20×10^{-5}
24	5	2.46×10^{-5}	45	1.20×10^{-7}
26	6	1.79×10^{-4}	46	3.70×10^{-6}
28	18	1.24×10^{-6}	47	2.30×10^{-7}
30	19	7.78×10^{-5}	48	4.40×10^{-6}
32	20	2.69×10^{-4}	50	2.80×10^{-5}
34	21	3.30×10^{-7}	51	5.00×10^{-6}
36	22	5.58×10^{-8}	52	6.00×10^{-7}
38	23	8.90×10^{-7}	53	1.70×10^{-7}
40	24	1.10×10^{-7}	54	2.20×10^{-6}
42	25	3.59×10^{-4}	55	1.30×10^{-8}
44	27	8.59×10^{-7}	56	1.10×10^{-7}
46	30	9.00×10^{-8}	57	3.00×10^{-7}
48	31	2.80×10^{-7}	59	7.50×10^{-6}
50	32	1.20×10^{-7}	60	7.50×10^{-7}
52	33	1.50×10^{-5}	61	1.50×10^{-7}
54	34	3.59×10^{-4}	63	1.30×10^{-6}
56	37	6.60×10^{-7}	64	8.80×10^{-7}
58	38	7.90×10^{-6}	73	1.30×10^{-3}

2. Therapeutic Action to Nephritis.

40 When puromycin aminonucleoside is administered to rats, symptoms similar to protein-rich urine, hypoproteinemia, hyperlipemia, nephrotic syndrome, etc. result and, therefore, rats which were administered with puromycin aminonucleoside have been used as pathological model animals for nephritis. A method by Endo, et al. [Sogo Rinsho, vol.38, no.5, page 821 (1989)] was somewhat modified and used as a test method here. Thus, a solution of puromycin aminonucleoside was dissolved in a physiological saline solution and administered just once into the tail vein of male rats (SD strain) of about 200 g body weight at a dose of 100 mg/kg (the initial or zero-th day).

45 The compound to be tested was dissolved in a physiological saline solution and was given orally for consecutive five days from the zero-th day at a dose of 50 mg/kg each. After 24 hours, the accumulated urine was collected and the amount of urine and that of protein in the urine were measured. Blood was collected on the tenth day and total protein in serum, creatinine in serum and urea nitrogen were measured.

50 Examples of the results are given in Tables 2 and 3.

TABLE 2

Compound to be tested	Urea nitrogen (mg/dl)	Total protein in serum (g/dl)	Serum creatinine (mg/dl)
(Normal)	17.6	7.90	0.38
(Control)	40.0	7.08	0.85
Compound 27	31.9	7.44	0.71

TABLE 3

Days	Amount of protein in urine (mg/day)	
	Control	Compound 27
1st day	0	0
2nd	32.0	29.2
3rd	44.7	33.8
4th	173.4	122.5
5th	522.5	399.8
6th	704.1	443.7
7th	826.5	558.7
8th	811.6	595.6
9th	814.0	593.5
10th	684.0	528.6

3. Inhibitory Action against Generation of Activated Oxygen

Human peripheral polymorphonuclear leukocytes (PML, 2×10^6 cells) prepared in a conventional method, bovine heart cytochrome C type-III (75 nmol), cytocharasin (5 μ g) and the tested drug were mixed with HEPES-buffered saline solution (final amount 1 ml) and incubated for 5 minutes at 37°C. N-Formyl-methionyl-leucyl-phenylalanine (FMLP) was added (final concentration 10^{-7} M) and incubated for 5 minutes. Immediately after incubation, the reaction mixture was centrifuged at 4°C, and then the absorbance of the supernatant at 550 nm was measured with a spectrophotometer.

An excess amount of bovine liver superoxide dismutase (SOD) was added to said reaction mixture and the absorbance of the supernatant was also measured in the same manner as above as a blank value.

A portion of adenosinedeaminase (ADA) may be removed during the preparation of human peripheral PML. Therefore, the inhibitory action of the tested drug against the generation of superoxide was measured in the same manner as mentioned above, adding 0.02 units of bovine liver ADA with human peripheral PML.

The inhibition of the superoxide generation was calculated by the following equation, and examples of the results are given in Table 4.

45

$$\text{Inhibition (\%)} = \left(1 - \frac{\text{Drug A}_{550} - \text{SOD A}_{550}}{\text{Control A}_{550} - \text{SOD A}_{550}} \right) \times 100$$

50

55

TABLE 4

5	Without ADA		With ADA	
	Compound 27 (M)	Inhibition	Compound 27 (M)	Inhibition
10	1×10^{-3}	100%	1×10^{-4}	88%
	1×10^{-4}	79%	1×10^{-5}	77%
	1×10^{-5}	22%	1×10^{-6}	34%
	1×10^{-6}	2%	1×10^{-7}	10%

4. Suppressive Action against Ischemic Edema

15 The right hind paw of ICR-strain male mice (11 weeks old) was fastened by a rubber band to stop blood circulation in the paw for 20 minutes, and then the rubber band was removed to recover blood circulation. The tested drug was administered intravenously before said treatment. Each of the right and left hind paws was weighed and the suppressive action was measured according to the weight difference between treated and untreated paws. Examples of the results are given in Table 5.

20

TABLE 5

25	Tested Drug (10 mg/kg)	Paw Weight (mg)		Inhibition (%)
		Treated	Untreated	
	Control	314.8 ± 6.8	219.9 ± 6.2	-
	Compound 27	254.4 ± 6.6	223.8 ± 6.2	67.7
	Allopurinol	283.2 ± 6.3	222.9 ± 6.9	36.4

30

5. Effect on the Concentrations of Adenosine and Inosine

35 Human peripheral polymorphonuclear leukocytes (2×10^6 cells), cytocharasin (5 μ g) and the tested drug were mixed with HEPES-buffered saline solution (final amount 0.5 ml) and incubated for 5 minutes. N-Formyl-methionyl-leucyl-phenylalanine (FMLP) was added (final concentration 10^{-7} M) and incubated for 10 minutes. As a result of HPLC analyses of the reaction mixture, in the FMLP-treated group, the inosine peak increased compared with the control group. The compound 27 of the present invention was added to the FMLP-treated group, which resulted in a significant decrease of the inosine peak and increase of the adenosine peak as compared with a group wherein the compound of the present invention was not added.

40 As shown in the above results (cf. Table 1), the compounds of the present invention exhibit an excellent adenosinedeaminase inhibiting action. Adenosinedeaminase which is a metabolic enzyme for adenosine is inhibited whereby the adenosine concentration in tissues increases. Neutrophils produce activated oxygen when the tissue is in an ischemic state and adenosine inhibits said activated oxygen production and, in addition, it directly eliminates the produced activated oxygen. Further, it lowers the inosine concentration whereby it decreases the supply of hypoxanthine which is a substrate of the xanthine-xanthineoxidase system (which is one of the systems producing the activated oxygen).

45 Adenosinedeaminase inhibiting substances having an oxygen production inhibiting action and an eliminating action for an activated oxygen source as such show pharmacological actions such as the improvement of coronary and cerebral blood vessel circulation, prevention and therapy of renal diseases, anti-inflammatory activity, etc.

50 Further, as shown in Tables 2 and 3, the compounds of the present invention having adenosinedeaminase inhibiting action were evaluated by means of pharmacological experiments using pathological model animals for nephritis induced by puromycin aminonucleoside, taking protein in urine, total protein in serum, creatinine in serum, urea nitrogen, etc. as indexes whereby the therapeutic effect was clearly noted.

55 Consequently, the compounds of the present invention having adenosinedeaminase inhibiting action are useful as pharmaceuticals for prevention and therapy of various kinds of diseases such as ischemic heart

diseases (e.g. angina pectoris, myocardial infarction and arrhythmia), diseases caused by cerebrovascular disorders (e.g. cerebral hemorrhage, cerebral infarction, cerebral apoplexy and after-effects thereof and cerebral arteriosclerosis), renal diseases (e.g. nephritis and renal failure) and allergic diseases (e.g. asthma, allergic rhinitis, allergic conjunctivitis, urticaria and rheumatism). Moreover, they are very useful as 5 pharmaceuticals for prevention and therapy of post-operative complicated diseases because they inactivate activated oxygen which is generated in ischemic areas during the recirculation of blood after operations.

Adenosine analogs such as 3'-deoxyadenosine and xylosyladenine (anticancer drugs) and arabinosyladenine (exhibiting antiherpes activity) are easily deaminated by adenosinedeaminase in vivo and thus become inactive. Accordingly, when the compounds of the present invention having adenosinedeaminase inhibiting action are administered before or together with administration of the above-10 mentioned anticancer drugs or antiviral drugs, an effect of inhibiting the decrease in action of such adenosine analogous anticancer and antiviral drugs can be expected as well.

Adenosine itself has many pharmacological activities such as a cardiovasodilating or platelet-aggregation inhibiting activity. Thus, it is used as a drug improving blood circulation to treat heart failure, myocardial 15 infarction, etc. Adenosine is metabolized by adenosinedeaminase, and is consequently inactivated. Accordingly, in the same way as above, when the compounds of the present invention are administered before or together with the administration of adenosine, the instant compounds may inhibit the decrease of said activities of adenosine. It is therefore advantageous to administer adenosine in combination with a compound of the present invention.

20 The compounds of the present invention can be made into pharmaceuticals by combining with a suitable pharmaceutically acceptable carrier or diluent for medical use and can be made into pharmaceutical preparations by any of conventional methods giving solid, semisolid, liquid or gaseous forms for oral or parenteral administration. In manufacturing such preparations, the compounds of the present invention may be used in a form of pharmaceutically acceptable salts thereof or the compound of the present invention 25 may be used either solely or jointly in a form of a suitable combination. Alternatively, the compound may be compounded with other pharmaceutically active components.

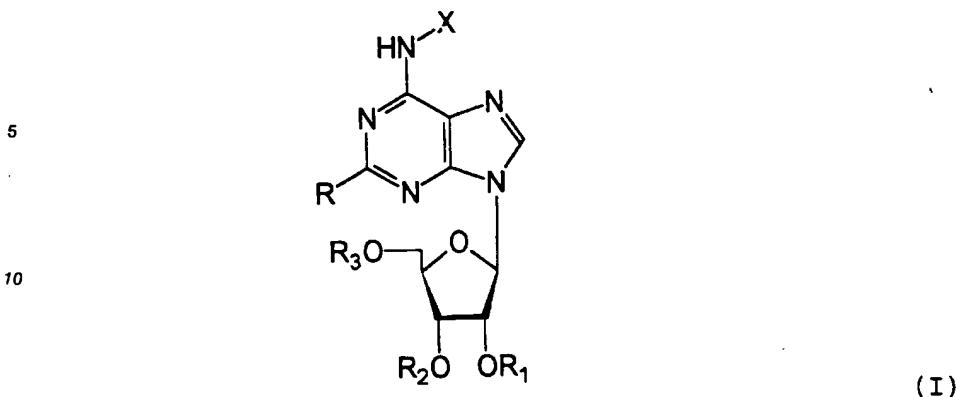
In the case of preparations for oral use, the compound of the present invention may be made into tablets, diluted powders, granules or capsules with or without suitable additives such as conventional fillers (e.g. lactose, mannitol, corn starch and potato starch) as well as binders (e.g. crystalline cellulose, cellulose 30 derivatives, gum arabic, corn starch and gelatin), lubricants (e.g. talc and magnesium stearate), extenders, moisturizers, disintegrating agents, preservatives, perfumes, etc. Alternatively, the compound of the present invention may be mixed with base material of fat/oil type (e.g. cacao butter), an emulsifying base material, water-soluble base material (e.g. Macrogel), a hydrophilic base material, etc. to give an ointment.

In the case of injections, the compound may be made into a solution or a suspension in aqueous 35 solvents or nonaqueous ones such as distilled water for injection, physiological saline liquid, Ringer solution, plant oil, synthetic fatty acid glycerides, higher fatty acid esters, propylene glycol, etc. Further, depending upon the state of the patient, or the type of the disease, the compound may be made into other preparation forms which are most suitable for the therapy such as inhalants, aerosols, ointments, poultices, eye drops, etc. The desired dosage of the compound of the present invention may vary depending upon the patient to 40 be treated, the preparation form, method of administration, period for administration, etc., though, in general, 0.1 to 5,000 mg or, preferably, 0.2 to 3,000 mg per day may be given to an adult by oral route for achieving the desired effect.

In the case of parenteral administrations such as injections, there are many cases where a lower dose than the above by oral administration to an extent of from 1/3 to 1/10 is desired because of the influence of 45 absorption, etc.

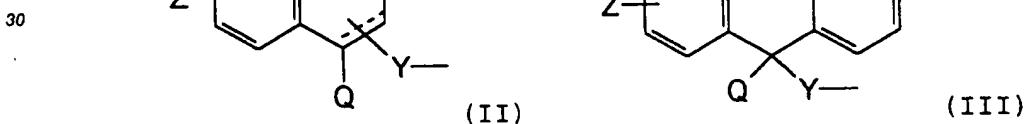
Claims

1. The use of a compound represented by the following general formula (I) or a pharmaceutically acceptable salt thereof



15

wherein each of R₁, R₂ and R₃ which may be same or different is hydrogen or alkyl;
 R is hydrogen, alkyl, alkenyl, alkynyl, hydroxyalkynyl, alkoxy, phenyl, hydroxy, amino, alkylamino, phenylamino or halogen;
 20 X is hydrogen, alkyl, alkynyl, allyl, methallyl, cycloalkyl, an alkyl group being substituted with one or more hydroxy groups, phenyl (which may be substituted with one or more halogen atoms, lower alkyl groups, lower alkoxy groups and/or trifluoromethyl groups), an alkyl group being substituted with one or more phenyl groups (which may be substituted with one or more halogen atoms, lower alkyl groups, lower alkoxy groups and/or trifluoromethyl groups), bicycloalkyl, naphthylalkyl, acenaphthylenealkyl or
 25 a group represented by the following general formulae (II) or (III)



35

wherein Z is hydrogen, hydroxy or lower alkoxy;
 Q is hydrogen or hydroxy;
 A is -CH₂-, -O-, -S- or a single bond;
 Y is -(CH₂)_n- or a single bond with n being an integer of 1 to 3;
 40 a dashed line means a single bond or a double bond;
 and wherein at least one of R₁, R₂ and R₃ is alkyl,
 for the preparation of a medicament which is an adenosinedeaminase inhibitor.

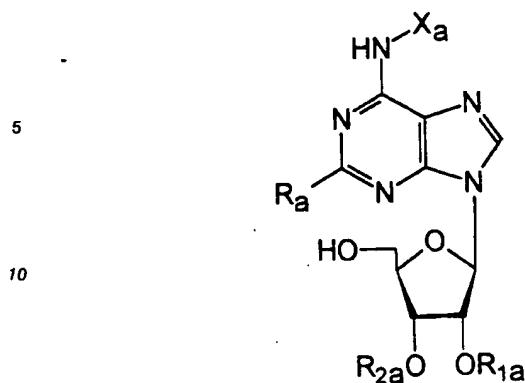
2. The use according to claim 1 wherein R₃ is hydrogen.

45 3. The use according to claim 2 wherein X is hydrogen.

4. The use according to claim 2 wherein X is an alkyl group having 1 to 3 carbon atoms.

50 5. Adenosine derivatives represented by the following general formula (Ia) or pharmaceutically acceptable salts thereof

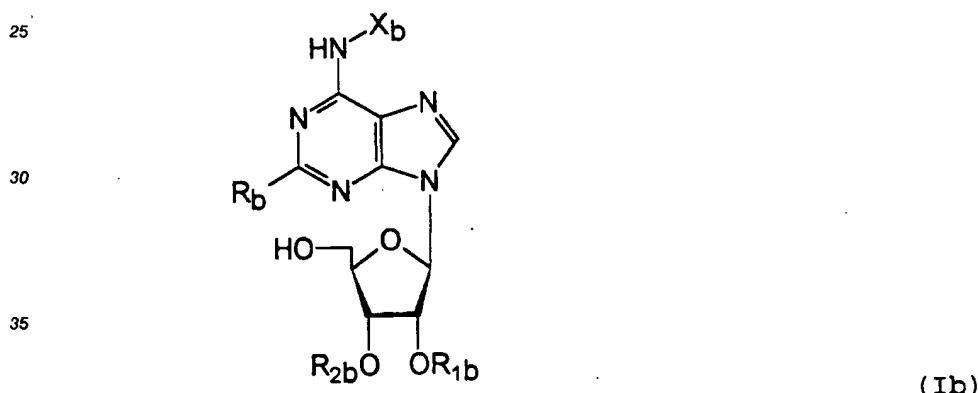
55



wherein each of R_{1a} , R_{2a} and X_a may be same or different and is hydrogen or alkyl; R_a is alkyl having more than 6 carbon atoms, alkenyl, alkynyl, hydroxyalkynyl, alkoxy, phenyl, hydroxy, alkylamino or phenylamino; and at least one of R_{1a} and R_{2a} is alkyl.

20

6. Adenosine derivatives represented by the following general formula (Ib) or pharmaceutically acceptable salts thereof,



40

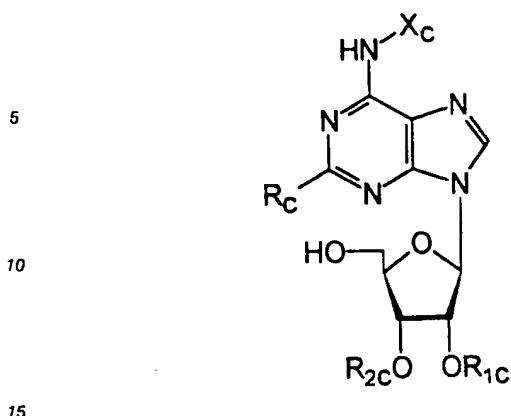
wherein each of R_{1b} and R_{2b} may be same or different and is alkyl; R_b and X_b is hydrogen or alkyl; and at least one of R_b and X_b is alkyl.

45

7. Adenosine derivatives represented by the following general formula (Ic) or pharmaceutically acceptable salts thereof,

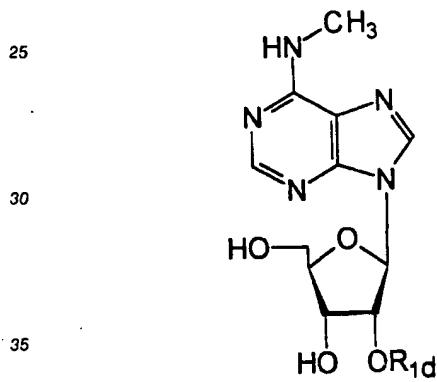
50

55



wherein each of R_{1c} , R_{2c} and X_c may be same or different and is hydrogen or alkyl; R_c is bromo or iodo; and at least one of R_{1c} and R_{2c} is alkyl.

20 8. Adenosine derivatives represented by the following general formula (Id) or pharmaceutically acceptable salts thereof



wherein R_{1d} is an alkyl group having more than 2 carbon atoms.

40 9. Pharmaceutical composition comprising at least one compound of formulae (Ia), (Ib), (Ic) or (Id) or a pharmaceutically acceptable salt thereof according to anyone of claims 5 to 8 in combination with a pharmaceutically acceptable carrier or diluent.

45 10. The use of a compound of formulae (Ia), (Ib), (Ic) or (Id) or a pharmaceutically acceptable salt thereof according to anyone of claims 5 to 8 for the preparation of a medicament which is an adenosinedeaminase inhibitor.

50 11. The use according to anyone of claims 1-4 and 10 wherein the medicament is for the prevention or therapy of ischemic heart diseases, diseases caused by cerebrovascular disorders, renal diseases and allergic diseases and of post-operative complicated diseases involved with activated oxygen which is generated in ischemic areas during the recirculation of blood after operations.

55 12. A medicament, comprising a combination of
 (A) a compound of formula (I) as defined in claim 1 which acts as an adenosinedeaminase inhibitor and
 (B) adenosine or a therapeutically useful adenosine analogous drug which is different from the compounds of formula (I),

as active ingredients.

13. The medicament of claim 12, wherein the adenosine analogous drug is an anticancer and/or an antiviral drug.

5

14. The medicament of claim 13, wherein the anticancer drug is 3'-deoxyadenosine or xylosyladenine and the antiviral drug is arabinosyladenine.

10

15

20

25

30

35

40

45

50

55



Europäisches Patentamt
European Patent Office
Office européen des brevets



⑪ Publication number:

0 601 322 A3

⑫

EUROPEAN PATENT APPLICATION

⑬ Application number: 93117408.0

⑮ Int. Cl.5: A61K 31/70, C07H 19/16

⑯ Date of filing: 27.10.93

⑭ Priority: 27.10.92 JP 312764/92
27.10.92 JP 312765/92

1-2, Hiranomachi Nichome
Chuo-ku, Osaka (JP)

⑯ Date of publication of application:
15.06.94 Bulletin 94/24

⑰ Inventor: Yamada, Toshio, c/o Inst. of
Bio-Active Science
Nippon Zoki Pharma. Co.,Ltd.,
442-1, Klnashi
Yashiro-cho, Kato-gun, Hyogo (JP)

⑯ Designated Contracting States:
AT BE CH DE DK ES FR GB GR IE IT LI LU MC
NL PT SE

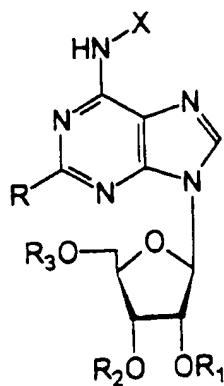
⑯ Date of deferred publication of the search report:
12.10.94 Bulletin 94/41

⑰ Representative: Hansen, Bernd, Dr.
Dipl.-Chem. et al
Hoffmann, Eitlie & Partner,
Patentanwälte,
Arabellastrasse 4
D-81925 München (DE)

⑰ Applicant: NIPPON ZOKI PHARMACEUTICAL
CO., LTD.

⑯ Adenosine deaminase inhibitor.

⑯ The use of a compound represented by the following general formula (I) or a pharmaceutically acceptable salt thereof



(I)

EP 0 601 322 A3

wherein each of R₁, R₂ and R₃ which may be same or different is hydrogen or alkyl; and R and X have the meanings given in the specification, for the preparation of a medicament which is an adenosine deaminase inhibitor is disclosed. Some of the compounds of formula (I) are novel compounds. They may be used for the making of a medicament which is for the prevention or therapy of ischemic heart diseases, diseases caused by cerebrovascular disorders, renal diseases and allergic diseases and of post-operative complicated diseases involved with activated oxygen which is generated in ischemic areas during the recirculation of blood after operations.



European Patent
Office

EUROPEAN SEARCH REPORT

Application Number
EP 93 11 7408

DOCUMENTS CONSIDERED TO BE RELEVANT									
Category	Citation of document with indication, where appropriate, of relevant passages	Relevant to claim	CLASSIFICATION OF THE APPLICATION (Int.Cl.5)						
X	EP-A-0 269 574 (NIPPON ZOKI PHARMACEUTICAL CO., LTD.) * page 3, line 31 - page 4, line 29; examples 1, and, 2 * ---	1-14	A61K31/70 C07H19/16						
X	BIOCHEMICAL PHARMACOLOGY, vol.41, no.11, 1991, NEW YORK pages 1615 - 1624 J.G.NIEDZWICKI ET AL. 'Structure-Activity Relationship of Ligands of Human Plasma Adenosine Deaminase2.' * abstract * * page 1619, line 15; example LXXXII * ---	1-14							
Y	WO-A-92 05177 (RHONE-POULENC RORER INTERNATIONAL) * claim 1 * ---	1-14							
X	EP-A-0 490 818 (SANDOZ-PATENT-GMBH) * abstract * ---	1-14							
Y	EP-A-0 378 518 (NIPPON ZOKI PHARMACEUTICAL CO., LTD.) * abstract * ---	1-14							
Y	WO-A-88 03147 (WARNER-LAMBERT COMPANY) * abstract * ---	1-14							
Y	EP-A-0 181 129 (WARNER-LAMBERT COMPANY) * abstract * ---	1-14							
Y	EP-A-0 219 876 (YAMASA SHPYU K.K.) * abstract * ---	1-14							
		-/-							
<p>The present search report has been drawn up for all claims</p> <table border="1"> <tr> <td>Place of search</td> <td>Date of completion of the search</td> <td>Examiner</td> </tr> <tr> <td>THE HAGUE</td> <td>3 August 1994</td> <td>Scott, J</td> </tr> </table> <p>CATEGORY OF CITED DOCUMENTS</p> <p>X : particularly relevant if taken alone Y : particularly relevant if combined with another document of the same category A : technological background O : non-written disclosure P : intermediate document</p> <p>T : theory or principle underlying the invention E : earlier patent document, but published on, or after the filing date D : document cited in the application I : document cited for other reasons & : member of the same patent family, corresponding document</p>				Place of search	Date of completion of the search	Examiner	THE HAGUE	3 August 1994	Scott, J
Place of search	Date of completion of the search	Examiner							
THE HAGUE	3 August 1994	Scott, J							



GEBÜHRENPLICHTIGE PATENTANSPRÜCHE

Die vorliegende europäische Patentanmeldung enthielt bei ihrer Einreichung mehr als zehn Patentansprüche.

- Alle Anspruchsgebühren wurden innerhalb der vorgeschriebenen Frist entrichtet. Der vorliegende europäische Recherchenbericht wurde für alle Patentansprüche erstellt.
- Nur ein Teil der Anspruchsgebühren wurde innerhalb der vorgeschriebenen Frist entrichtet. Der vorliegende europäische Recherchenbericht wurde für die ersten zehn sowie für jene Patentansprüche erstellt für die Anspruchsgebühren entrichtet wurden,
nämlich Patentansprüche:
- Keine der Anspruchsgebühren wurde innerhalb der vorgeschriebenen Frist entrichtet. Der vorliegende europäische Recherchenbericht wurde für die ersten zehn Patentansprüche erstellt.

MANGELNDE EINHEITLICHKEIT DER ERFINDUNG

Nach Auffassung der Recherchenabteilung entspricht die vorliegende europäische Patentanmeldung nicht den Anforderungen an die Einheitlichkeit der Erfindung; sie enthält mehrere Erfindungen oder Gruppen von Erfindungen,
nämlich:

1. Patentansprüche 1-3,5-35: Bandage mit Pelotte (50) sowie Bandage mit Pelotte (150).
2. Patentanspruch 4: Bandage mit Pelotte (60,60').

- Alle weiteren Recherchengebühren wurden innerhalb der gesetzten Frist entrichtet. Der vorliegende europäische Recherchenbericht wurde für alle Patentansprüche erstellt.
- Nur ein Teil der weiteren Recherchengebühren wurde innerhalb der gesetzten Frist entrichtet. Der vorliegende europäische Recherchenbericht wurde für die Teile der Anmeldung erstellt, die sich auf Erfindungen beziehen,
für die Recherchengebühren entrichtet worden sind,
nämlich Patentansprüche:
- Keine der weiteren Recherchengebühren wurde innerhalb der gesetzten Frist entrichtet. Der vorliegende europäische Recherchenbericht wurde für die Teile der Anmeldung erstellt, die sich auf die zuerst in den Patentansprüchen erwähnte Erfindung beziehen,
nämlich Patentansprüche 1-3,5-35



European Patent
Office

EUROPEAN SEARCH REPORT

Application Number
EP 93 11 7408

DOCUMENTS CONSIDERED TO BE RELEVANT			
Category	Citation of document with indication, where appropriate, of relevant passages	Relevant to claim	CLASSIFICATION OF THE APPLICATION (Int.Cl.5)
Y	<p>BIOCHEMISTRY, vol.15, no.13, 1976 pages 2735 - 2740</p> <p>Z.KAZIMIERCZUK ET AL. 'Methylation of Adenosine in Strongly Alkaline Medium: Preparation and Properties of O'-Methyl Derivatives of Adenosine and N6-Methyladenosine.'</p> <p>* the whole document *</p> <p>---</p>	1-14	
Y	<p>NUCLEOSIDES AND NUCLEOTIDES, vol.10, no.8, 1991 pages 1677 - 1692</p> <p>G.MAURY ET AL. 'Inhibition and Substrate Specificity of Adenosine Deaminase. 2',3'- and/or 5'-Substituted Adenine Nucleoside Derivatives.'</p> <p>* the whole document *</p> <p>----</p>	1-14	
			TECHNICAL FIELDS SEARCHED (Int.Cl.5)
<p>The present search report has been drawn up for all claims</p>			
Place of search EPO FORM 1501/02/22 (PC/CON)	Date of completion of the search 3 August 1994	Examiner Scott, J	
CATEGORY OF CITED DOCUMENTS		<p>T : theory or principle underlying the invention E : earlier patent document, but published on, or after the filing date D : document cited in the application L : document cited for other reasons & : member of the same patent family, corresponding document</p>	
<p>X : particularly relevant if taken alone Y : particularly relevant if combined with another document of the same category A : technological background O : non-written disclosure P : intermediate document</p>			



European Patent
Office

EP 93 11 7408 -B-

LACK OF UNITY OF INVENTION

The Search Division considers that the present European patent application does not comply with the requirement of unity of invention and relates to several inventions or groups of inventions, namely:

1. Claims : 1-3,9-14 in part :

The use of compounds of formula (I)
claim 1, where R1=alkyl, R2=H, R3=H,
R=H and X=H as adenosine deaminase
inhibitors, pharmaceutical
compositions containing them, their
use, and medicaments containing them.

2. Claims : 1,2,4,5,9-14 in part : 8 wholly :

The use of compounds of formula (I)
claim 1, where R1=alkyl, R2=H, R3=H,
R=H and X is other than H as adenosine
deaminase inhibitors, compounds of
formula (Ia), claim 5, pharmaceutical
compositions containing them, their
use, and medicaments containing them.

3. Claims : 1-7,9-14 in part :

The use of compounds of formula (I)
claim 1, where R1=alkyl, R2=H, R3=H,
and R is other than H as adenosine
deaminase inhibitors, compounds of
formula (Ia), claim 5; (Ib), claim 6;
(Ic), claim 7, pharmaceutical
compositions containing them, their
use, and medicaments containing them.

4. Claims : 1-7,9-14 in part :

The use of compounds of formula (I),
claim 1, where two or three of R1,R2,
R3 are alkyl as adenosine deaminase
inhibitors, compounds of formula (Ia),



European Patent
Office

EP 93 11 7408 -B2-

LACK OF UNITY OF INVENTION

The Search Division considers that the present European patent application does not comply with the requirement of unity of invention and relates to several inventions or groups of inventions, namely:

claim 5; (Ib), claim 6; (Ic), claim 7,
pharmaceutical compositions containing
them, their use, and medicaments
containing them.

5. Claims : 1,9-14 in part :

The use of compounds of formula (I),
claim 1, where R1=H, R2=H, R3=alkyl as
adenosine deaminase inhibitors
pharmaceutical compositions containing
them, their use, and medicaments
containing them.

6. Claims : 1-7,9-14 in part :

The use of compounds of formula (I),
claim 1, where R1=H, R2=alkyl, R3=H as
adenosine deaminase inhibitors,
compounds of formula (Ia), claim 5;
(Ib), claim 6; (Ic), claim 7,
pharmaceutical compositions containing
them, their use, and medicaments
containing them.

The subject-matter searched and proposed for search defines solely the scope of the subject for the purpose of search and may not as defined satisfy the requirement of Article 123(2), EPC, with regard to possible granting of such subject-matter.

Since only the first subject-matter has been searched, then the search division reserves the right to formulate further non-unities should the necessity arise.

**This Page is Inserted by IFW Indexing and Scanning
Operations and is not part of the Official Record**

BEST AVAILABLE IMAGES

Defective images within this document are accurate representations of the original documents submitted by the applicant.

Defects in the images include but are not limited to the items checked:

- BLACK BORDERS**
- IMAGE CUT OFF AT TOP, BOTTOM OR SIDES**
- FADED TEXT OR DRAWING**
- BLURRED OR ILLEGIBLE TEXT OR DRAWING**
- SKEWED/SLANTED IMAGES**
- COLOR OR BLACK AND WHITE PHOTOGRAPHS**
- GRAY SCALE DOCUMENTS**
- LINES OR MARKS ON ORIGINAL DOCUMENT**
- REFERENCE(S) OR EXHIBIT(S) SUBMITTED ARE POOR QUALITY**
- OTHER:** _____

IMAGES ARE BEST AVAILABLE COPY.

As rescanning these documents will not correct the image problems checked, please do not report these problems to the IFW Image Problem Mailbox.